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EFFECTS OF FEEDING BEHAVIOR IN COPEPOD *Oithona similis* WITH INTAKE OF PHYTOPLANKTON

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Abstract

Copepods are usually the major component of marine metazooplankton and the key link from primary producers to fish production in pelagic food webs. Copepod feeding behavior differs between life stages of the same species and similarly, the effect of environmental factors would be different depending on the developmental stage. *Oithons similis* were collected at a station in Dona Paula Bay (15° 27' N, 73°N, 48' E), west coast of India. These copepods after separation were maintained in 500 ml beaker containing 0.22µm filtered sea water with algae, *Dunaiella tertiolecta*, *Isochrysis galbana* and *Chaetoceros calcitrans*. All experiments were performed at the same light and temperature (25 ± 2°C) conditions to the stock culture. After 24 hours wells were checked for the release of nauplii by gravid females. The nauplii were picked individually and placed in 24 well plates containing different food organisms. During the transfer observations were made for mortality and molt in each well. When a molt was observed, it indicates that nauplii or copepodite has moved to the next stage. The size was measure as total length using a binocular upright microscope. In the present study, the naupliar and copepodite development showed varying effect on their development and also it affected the size of the individual stages during development. The rate of survival was also maximum when reared with natural phytoplankton cells than compared to single species pure culture of phytoplanktons. It was also to be noted that the cell concentration of natural phytoplankton cells was comparatively low than pure culture of phytoplankton raised in the laboratory.

Keywords: Feeding behavior, Copepod- *Oithons similis* and Phytoplankton.

1.Introduction

Copepod is one of the most important groups of the crustaceans, their own taxonomic class, "The copepod". Currently, more than 13,000 species of copepods are known to science, but that number probably represents less than a quarter of the extant species. Copepods vary in size significantly; early naupliar stages of copepods may be less than 0.1 mm in length while the largest adult parasite is over 3 cm. Copepods are

usually the major component of marine metazooplankton (Longhurst, 1985; Verity and Smetacek, 1996) and the main prey of most fish larvae (Conway *et al.*, 1991; Conway *et al.*, 1998). Importance of life-cycle studies as a basis for understanding copepod phylogeny and for the construction of a satisfactory classification system cannot be over emphasized. There are two main reasons for studying the life cycle of copepods.

First, from a taxonomic point of view, knowing larval development will help reveal the natural relationships among adult organisms. Second, in order to better understand their importance in the ocean's economy, it is necessary to know the organisms accurately, to distinguish

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all their developmental stages, and to ascertain the habits and requirements of each (Antonio Trujillo-Ortiz, 1986). Life cycles have been described for at least 106 copepod species, but almost all of the descriptions are incomplete, because it was difficult to collect all developmental stages of copepods in nature and to keep them in culture.

Copepod nauplii have been historically under sampled by conventional methods such as 200- μ m mesh plankton nets (Alcaraz, 1977; Calbet *et al.*, 2001; Gallienne and Robins, 2001). When appropriate fine meshes of plankton nets have been employed, nauplii frequently outnumbered late copepodites and adults by several orders of magnitude (Turner, 1982; Turner, 1994; Turner, 2004; Hansen *et al.*, 1999; Calbet *et al.*, 2001) and sometimes, they represent a higher fraction of biomass than late stages (Castellani *et al.*, 2007). Moreover, nauplii can ingest small nanoplankton (Berggreen *et al.*, 1988) and their specific ingestion rates can be three to four times higher than those of adults (White and Roman, 1992; Lonsdale *et al.*, 1996).

Copepod feeding behavior differs between life stages of the same species (Fernandez, 1979) and similarly, the effect of environmental factors would be different depending on the developmental stage. Besides feeding rates, the gross growth efficiency (GGE) is another important measurement for detailing the flow of material from prey to predator. For the application of ecological models, GGE of zooplankton is commonly assumed to be constant (median 30%) for all types of zooplankton and during all the cycle life (Straile, 1997). However, GGE may vary significantly depending on the developmental stage and food concentration (Jones *et al.*, 2002; Rey-Rassat *et al.*, 2002). Consequently, general patterns of copepod feeding and growth efficiencies based on late stages should not be extrapolated to naupliar ones. Among marine copepods, cyclopoids of the genus *Oithona* are considered the most abundant and ubiquitous copepods in the world's oceans (Gallienne and

Robins, 2001). However, knowledge about the feeding and ecology of these small copepods was very scarce compared to the vast number of experimental studies devoted to calanoid copepods (Paffenhofer, 1993; Turner, 2004; Saiz and Calbet, 2007). Some information in the literature about the feeding rates of adult stages of *Oithona* in the laboratory (Saiz *et al.*, 2003) and in the field (Nakamura and Turner, 1997; Lonsdale *et al.*, 2000; Gifford *et al.*, 2007; Castellani *et al.*, 2005). However, there are few references on feeding rates of *Oithona nauplii* (Henriksen *et al.*, 2007). The present study attempts to provide basic information concerning the development of *Oithona similis* with respect to different food organisms cultured in the laboratory and also natural phytoplankton.

2. Materials and Methods

2.1. Collection and separation of *Oithona similis*

The adult *Oithona similis* were collected at a station in Dona Paula Bay (15° 27' N, 73°N, 48' E), West coast of India by using a Heron-Tranton net. The study site exhibits a highly dynamic environmental and considerable tidal influence from a neap tide of 0.25 m to a spring tide of 2.5 m. The surface water temperatures range from 25 to 35°C. Salinity fluctuates from 10 psu during the South-west monsoon (June to September) to 35 psu during the summer months (February to May).

After collection of zooplankton, samples were immediately taken to the laboratory and maintained at 25°C. Sample was kept near to light, as *O. similis* show phototactic behavior, they get concentrated near the light, which is then easier to collect them for separation. Precipitated material (feces and dead phytoplankton and zooplankton) was siphoned out. Aliquots were taken with Pasteur pipettes and transferred in to 250 ml beaker in order to facilitate the identification of adults of *O. similis*.



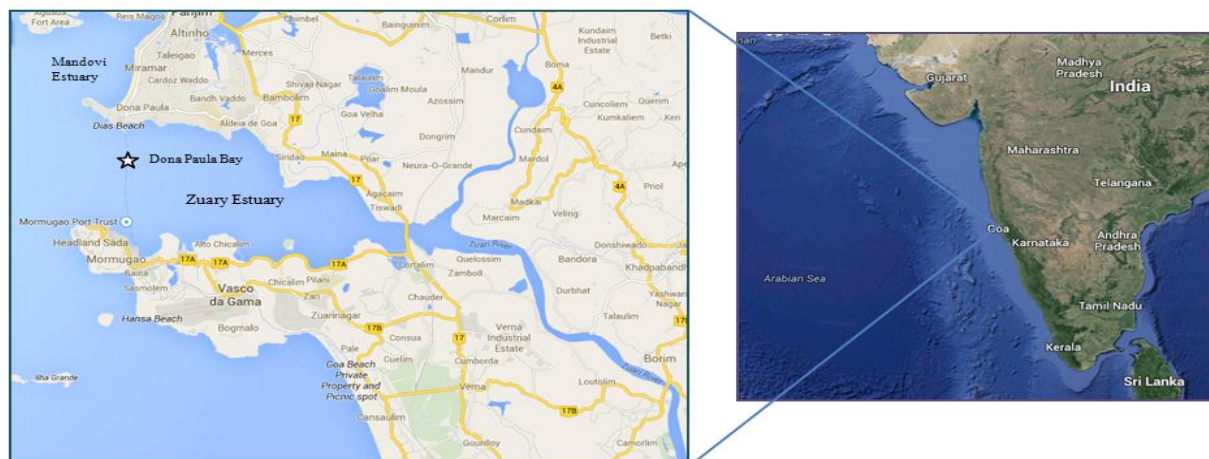


Figure – 1: Map providing the collection site for *O. similis* adults and phytoplankton

These copepods after separation were maintained in 500 ml beaker containing 0.22 μm filtered sea water with algae, *Dunaiella tertiolecta*, *Isochrysis galbana* and *Chaetoceros calcitrans*. All experiments were performed at the same temperature ($25 \pm 2^\circ\text{C}$) conditions to the stock culture.

2.2. Culturing algae

Dunaiella tertiolecta, *Isochrysis galbana* and *Chaetoceros calcitrans* were cultured as a feed for copepod. The algae were maintained in F/2 medium (F/2 medium Guillard and Ryther 1962).

2.3. Experimental setup

To examine the effect of food concentration on growth and development of *O. similis*, female *O. similis* carrying egg sacs were placed individually in six well multiwall plates containing 10 ml of different experimental food organisms and natural phytoplankton. After 24 hours wells were checked for the release of nauplii by gravid females. The nauplii were picked individually and placed in 24 wells plates containing different food organisms. At the end of each the nauplii and further the copepodites were transferred to fresh medium with food in different 24 wells plates. During the transfer, observations were made for mortality and molt in each well.

When a molt was observed, it indicates that nauplii or copepodite has moved to the next stage. The size was measure as total length using a binocular upright microscope.

2.4. Naupliar Stages

During postembryonic development, six naupliar stages are evident. The body is not significantly curved laterally. All naupliar stages are oval anteriorly, and narrow toward the caudal armature. There is an anteroventral pigment spot, generally red, also known as the naupliar eye. The posterior-inner part is tan, and the body is generally clear and translucent but slightly yellow-green. A small internal lipid body is usually present poster ventrally in most of the naupliar stages, and is clearly visible the labrum is clearly evident in all naupliar stages; in addition, there are short, thin setules in the labrum's inferior margin. After the first molt, the nauplii enlarge slightly, and the antennule, antenna and mandible become more specialized.

2.5. Copepodid and Adult Stages

The final naupliar stage (6th stage) metamorphoses into the first of copepodids. When the individual reaches copepodid stage I, as the direct result of a drastic change in structure, it is a miniature adult, except that it has only two pairs of functional swimming legs. A new pair of legs is added in each successive molt until copepodid IV.



From that stage until the adult form (copepodid VI), no new swimming legs are added; instead, the sexually modified fifth pair of legs develops in the adult form.

3. Results

3.1. Effect of different food organisms on development of *Oithona similis* *D. tertiolecta*

A food concentration of 3×10^5 cells/ml of *D.tertiolecta* was used to rear different naupliar and copepodite stages. While reared with this food the development was completed in 15+ 3 days from naupliar to adult stages (completion of copepodite stage). The 3rd and 5th naupliar stage took the longer duration to molt into the next stage, whereas among the copepodite, 4th copepodite stage took the longer duration 2+0.5 days to molt into the next stage.

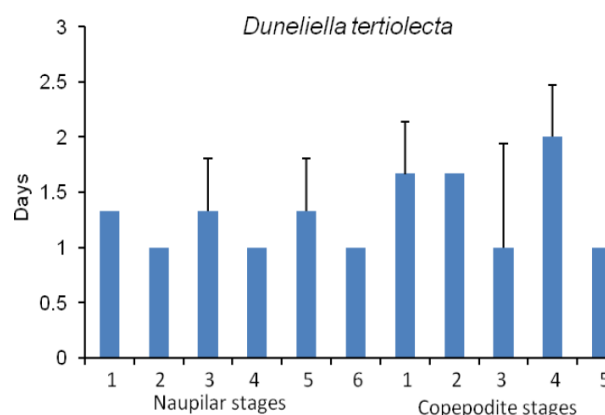


Figure – 2: Variations in the size of different naupliar and copepodite stages of *Oithona similis* feed with *Chaetoceros calcitrans*

3.3. *Dunaliella tertiolecta* with *Isochrysis galbana*

The mixed food cell concentration was 1.5×10^5 cells/ml of *Dunaliella tertiolecta* and 1.5×10^5 cells/ml of *Isochrysis galbana* was provided as food for *Oithona similis* naupliar and copepodite stages. The total development duration was 14+3 days. The maximum duration to molt into the next stage was for 1st nauplii and 1st copepodite stage which was 1.7+0.5 days and 2 days respectively.

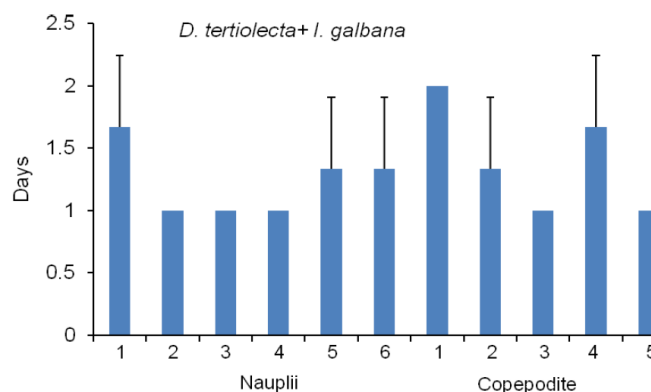


Figure – 3: Variations in the size of different naupliar and copepodite stages of *Oithona similis* feed with *Dunaliella tertiolecta* and *Isochrysis galbana*

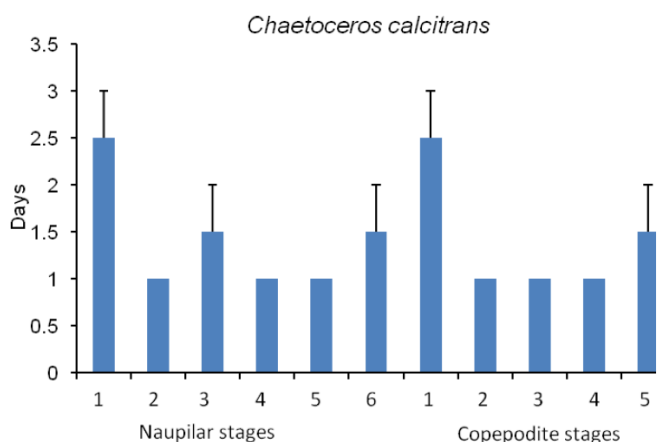


Figure – 1: Variations in the size of different naupliar and copepodite stages of *Oithona similis* feed with *D. tertiolecta*

3.2. *Chaetoceros calcitrans*

Food concentration 3×10^5 cells/ml of *Chaetoceros calcitrans* was used to rear naupliar and copepodite stages. The total development duration was 16+3 days. The maximum duration to molt into the next stage was for 2nd naupliar and 1st copepodite stage which was 2.5+1day respectively.



3.4. Natural Phytoplankton

The natural phytoplankton cells were provided for the development of different naupliar and copepodite stages. The cell concentration ranged from 150 to 250 cells/ml. The most common phytoplankton occurred in the sample was presented in Table - 1. The total development duration was minimum when fed with natural phytoplankton compared all other types of food. The total development duration was 12.5+1.5 days. The maximum duration to molt into the next stage was for 5th nauplii (1.5+0.5 days) and 4th and 5th copepodite stage which was 1.5+0.5 days respectively for both the stages.

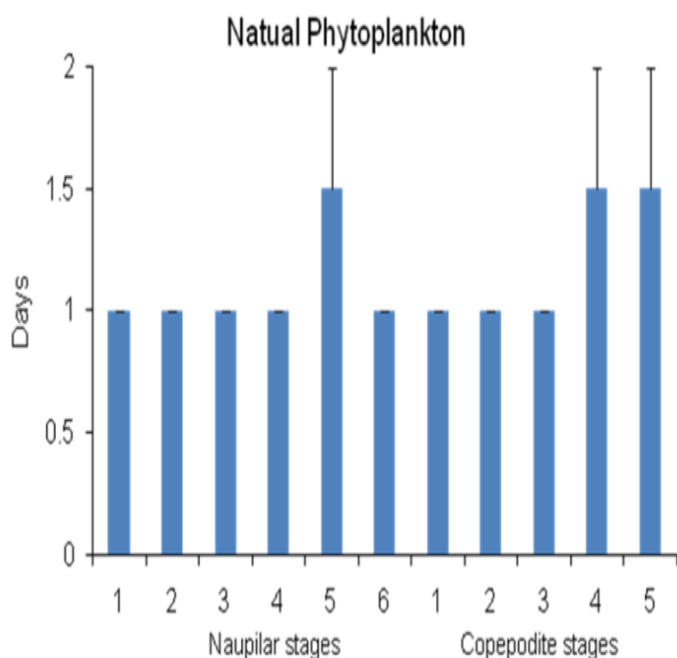


Figure – 4: Variations in the development duration of naupliar and copepodite stages of *Oithona similis* feed with natural phytoplankton

Table - 1: The common phytoplankton cells present in the natural sample provided as food for the naupliar and copepodite stages of *Oithona similis*.

S.no	Common phytoplankton species
01	<i>Asterionella</i> sp. (Diatom)
02	<i>Amphora</i> sp. (Diatom)
03	<i>Ceratium</i> sp. (Flagellate)
04	<i>Chaetoceros curvicaetus</i> (Diatom)
05	<i>Chaetoceros</i> sp. (Diatom)
06	<i>Coscinodiscus</i> sp. (Diatom)
07	<i>Cylindrus</i> sp. (Diatom)
08	<i>Gramatophora</i> sp. (Flagellate)
09	<i>Guinardia striata</i> (Diatom)
10	<i>Gyrodinium</i> sp.(Flagellate)
11	<i>Navicula</i> sp.(Diatom)
12	<i>Plurosigma</i> sp.(Diatom)
13	<i>Prorocentrum</i> sp. (Flagellate)
14	<i>Protopteridinium</i> sp. (Flagellate)
15	<i>Pseudo-nitzschia</i> sp.(Diatom)
16	<i>Skeletonema species</i> (Diatom)
17	<i>Thalassionema nitzschioides</i> (Diatom)

3.5. Effect of different food organisms on the size (total length) of naupliar and copepodite stages

The maximum size was attained by both nauplii and copepodite stages were when they were feed with natural phytoplankton and minimum size when they were feed with *Chaetocers calcitrans*. The total length of the 6th nauplii and 5th copepodite stage was 243 and 623 μ m when feed with natural phytoplankton and it was 200 and 580 μ m when feed with *Chaetocers calcitrans*.



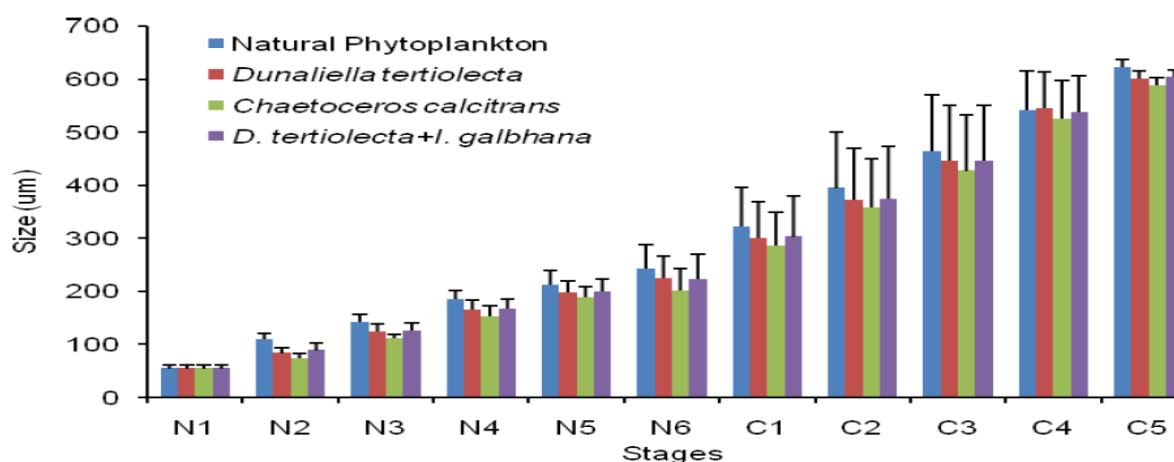


Figure - 5: Variations in the development duration of naupliar and copepodite stages of *Oithona similis* feed with *Dunaliella tertiolecta*, *Chaetoceros calcitrans* mixture of *Dunaliella tertiolecta* and *Isochrysis galbana*.

4. Discussion

This work describes the life cycle of the marine cyclopoida copepod *O. similis* reared in the laboratory. *Oithona* is one of the most abundant and cosmopolitan copepod groups throughout coastal and oceanic regions of tropical, temperate and polar waters. The life cycles of some *Oithona* spp. have been described previously, and most of these descriptions are incomplete. Identification of *Oithona* spp. is also difficult because these are small and the morphology of nauplii stages are similar. In the copepods most important factors affecting the development rates are temperature and food. The food concentration, variation in feeding rates, the adaptation of the feeding behaviors and energy demands of each copepod species and stages is different (Claudia *et al.*, 2008). When grown under conditions of unlimited food resources, development times from egg to adult are independent of adult body size. Males develop to adult more quickly than females. Differences in algal food quality and concentration, the copepod feeding behavior and developments stages are changed. Experimental studies have recently indicated that carnivorous feeding is not required to complete development stages and reproduction. In the present study the naupliar and copepodite development showed

varying effect on their development and also it affected the size of the individual stages during development. The rate of survival was also maximum when reared with natural phytoplankton cells than compared to single species pure culture of phytoplankton's. It is also to be noted that the cell concentration of natural phytoplankton cells was comparatively low than pure culture of phytoplankton raised in the laboratory.

The physiological condition of growing cells is a critical factor in the food selection process of zooplankton, and strongly influences the rate at which those growing cells are ingested. The copepod *Acartia tonsa* ingesting rates on faster growing cells of the diatom *Thalassiosira weissflogii* compared to ingested rates on slower growing cells of that species at the same cell concentration. (Cowles *et al.*, 1988). The development of nauplii is a function of environmental variables, food concentration and temperature. The *Oithona* spp. are ingesting different algal species, each species are feeding particular algae suggest that these differences may help to explain the ubiquity of *Oithona* spp. in oceanic environments. The differences in the different developmental rate can be attributed to varying levels of feeding depending upon the type of food given similar cell concentration. The lower



feeding can be interpreted as an adaptation to conserve energy at low food concentrations because the energetic cost of collecting food at very low concentrations would not be compensated by the energetic gain. Feeding ingesting rates vary depending on copepod species but are also influenced by other variables such as the developmental stage and the size and quality of the prey (Rodrigo *et al.*, 2010). In the case of *O. similis*, feeding ingesting rates were observed in the functional responses under food concentrations (*D. tertiolecta*) in the 3×10^5 cells/ml for early nauplii. With varying food and concentration the developmental stages and life cycle are changed. Then given to food for *C. calcitrans* the same concentration of food for the *O. similis* developments stages are similar but decrease in body size, longer development duration. The size of the *C. calcitrans* is small compared to other food organisms provided in the experiment. This may be one of the reasons for the delayed development and smaller naupliar and copepodite stages.

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